PATENT APPLICATION

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re the Application of:

KILGER et al — Group Art Unit: 1656

Application No.: New Application — Examiner: Horlick, K.

Filed: January 8, 2001 — Attorney Dkt. No.: 101614-00009

For: METHOD FOR THE UNCOUPLED, DIRECT, EXPONENTIAL AMPLIFICATION AND SEQUENCING OF DNA MOLECULES WITH THE ADDITION OF A SECOND THERMOSTABLE DNA POLYMERASE AND ITS APPLICATION

PRELIMINARY AMENDMENT

Commissioner for Patents
Washington, D.C. 20231

January 8, 2001

Sir:

Prior to initial examination of the application, please amend the above-identified application as follows:

IN THE CLAIMS:

Cancel claims 1-33 without prejudice and insert the following new claims:

-- 34. (New) A process for simultaneously amplifying and sequencing a nucleic acid molecule, comprising the steps of:

a) denaturing a double stranded DNA molecule to obtain two complementary single stranded DNA molecules;

b) contacting said two complementary single stranded DNA molecules with two primers to obtain two primer containing single stranded DNA molecules, wherein one of the two primers is complementary to one of the two complementary single stranded

DNA molecules and the other of the two primers is complementary to the other of the two complementary single stranded DNA molecules;

c) contacting said two primer containing single stranded DNA molecules with: (i) a set of chain elongating nucleotides, (ii) at least one chain terminating nucleotide, (iii) a first DNA polymerase; and (iv) a second DNA polymerase, which has a higher affinity towards the chain terminating nucleotide relative to the first polymerase, so that polymerization by the first polymerase results in amplification and polymerization by the second polymerase results in the formation of chain terminated fragments;

d) repeating steps a) through c) for an appropriate number of times to obtain chain terminating fragments for detection; and

e) detecting the chain terminated fragments by a detection means and aligning the fragments to determine the sequence of the nucleic acid molecule.

35. (New) A process of claim 34, wherein the first and second polymerases are thermostable DNA polymerases.

36. (New) A process of claim 34, wherein the set of chain-elongating nucleotides is i) at least one deoxyadenosine triphosphate; ii) at least one deoxyguanosine triphosphate; iii) at least one deoxycytidine triphosphate; and iv) at least one thymidine triphosphate.

37. (New) A process of claim 36, wherein the deoxyadenosine and/or the deoxyguanosine is an N7-deazapurine nucleotide.

38. (New) A process of claim 34, wherein the chain terminating nucleotide is selected from the group consisting of 2',3'-dideoxyadenosine triphosphate, 2',3'-dideoxyguanosine triphosphate, 2',3'-dideoxycytidine triphosphate, and 2',3'-dideoxythymidine triphosphate.

39. (New) A process of claim 34, wherein the detection means is a separation means in conjunction with a visualization means selected from the group consisting of: colorimetry, fluorimetry, chemiluminescence and radioactivity, wherein the separation means is polyacrylamide gel electrophoresis.

40. (New) A process of claim 34, wherein the double stranded DNA molecule has been synthesized from RNA using a reverse transcriptase.

41. (New) A process of claim 34, wherein the primer is mobility modified and the amplified and chain terminated fragments are detected by electrophoresis.

42. (New) A process for simultaneously amplifying and sequencing a single stranded nucleic acid molecule, comprising the steps of:

a) contacting the single stranded DNA molecule with: (i) a primer that can hybridize to the single stranded DNA molecule, (ii) a set of chain elongating nucleotides, (iii) at least one chain terminating nucleotide, (iv) a first DNA polymerase; and (v) a second DNA polymerase, which has a higher affinity towards the chain terminating nucleotide relative to the first polymerase, so that polymerization by the first polymerase results in

amplification and polymerization by the second polymerase results in the formation of chain terminated fragments;

b) detecting the chain terminated fragments by a detection means; and

c) aligning the fragments to determine the sequence of the single stranded nucleic acid molecule.

43. (New) A process of claim 42, wherein the first and second polymerases are thermostable DNA polymerases.

44. (New) A process of claim 42, wherein the set of chain-elongating nucleotides is i) at least one deoxyadenosine triphosphate; ii) at least one deoxyguanosine triphosphate; iii) at least one deoxycytidine triphosphate; and iv) at least one thymidine triphosphate.

45. (New) A process of claim 44, wherein the deoxyadenosine and/or the deoxyguanosine is an N7-deazapurine nucleotide.

46. (New) A process of claim 42, wherein the chain terminating nucleotide is selected from the group consisting of 2',3'-dideoxyadenosine triphosphate, 2',3'-dideoxyguanosine triphosphate, 2',3'-dideoxycytidine triphosphate, and 2',3'-dideoxythymidine triphosphate.

47. (New) A process of claim 42, wherein the detection means is a separation means in conjunction with a visualization means selected from the group consisting of:

colorimetry, fluorimetry, chemiluminescence and radioactivity, wherein the separation means is polyacrylamide gel electrophoresis.

48. (New) A process of claim 42, wherein the double stranded DNA molecule has been synthesized from RNA using a reverse transcriptase.

49. (New) A process of claim 42, wherein the primer is mobility modified and the amplified and chain terminated fragments are detected by electrophoresis.

50. (New) A kit for obtaining amplified and chain terminated nucleic acid molecules from a nucleic acid template comprising:(i) a set of chain elongating nucleotides, (ii) at least one chain terminating nucleotide, (iii) a first DNA polymerase; and (iv) a second DNA polymerase, which has a higher affinity towards at least one chain terminating nucleotide relative to the first polymerase.

51. (New) A kit of claim 50, wherein the first and second polymerases are thermostable DNA polymerases.

52. (New) A kit of claim 51, wherein the thermostable DNA polymerases are selected from the group consisting of: Taq DNA polymerase, AmpliTaq FS DNA polymerase, Thermo Sequenase, and Tth DNA polymerase.

53. (New) A kit of claim 50, wherein the set of chain-elongating nucleotides is i) at least one deoxyadenosine triphosphate; ii) at least one deoxyguanosine triphosphate; iii) at least one deoxycytidine triphosphate; and iv) at least one thymidine triphosphate.

54. (New) A kit of claim 53, wherein the deoxyadenosine and/or the deoxyguanosine is an N7-deazapurine nucleotide.

55. (New) A kit of claim 50, wherein the chain terminating nucleotide is selected from the group consisting of 2',3'-dideoxyadenosine triphosphate, 2',3'-dideoxyguanosine triphosphate, 2',3'-dideoxycytidine triphosphate, and 2',3'-dideoxythymidine triphosphate.

56. (New) A kit of claim 50, which additionally includes at least one primer.

57. (New) A kit of claim 56, wherein the primer is mobility modified and the amplified and chain terminated nucleic acid molecules are detected by electrophoresis.

58. (New) A kit of claim 50, which additionally comprises a reverse transcriptase.--

REMARKS

Claims 34-58 are pending.

Claims 34-58 are presented to support a request for an interference with Köster et al., U.S. Patent No. 5,928,906.

Claims 34-41 correspond to claims 33-38, 41 and 46, respectively, of Köster et al., U.S. Patent No. 5,928,906.

Claims 42-49 correspond to claims 17-22, 25 and 30, respectively, of Köster et al., U.S. Patent No. 5,928,906.

Claims 50-58 correspond to claims 1, 3-8, 13, and 16, respectively, of Köster et al., U.S. Patent No. 5,928,906 (herein after referred to as US '906).

The support for the limitations recited in claims 34-58 in the parent application, 09/357,166, and in the priority document is discussed in a Preliminary Amendment filed on July 26, 2000 during the prosecution of the parent application. A certified English translation of the priority document was filed in the grandparent application, 08/991,184. Applicants request that the Patent Office grant a priority date of December 20, 1996 for the invention according to claims 34-58.

Please charge any fee deficiency or credit any overpayment to Deposit Account No. 01-2300.

Respectfully submitted,

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